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The Effect of Mixing Time and Mixing Sequence during Processing on the Physicochemical and Sensory Properties of Keropok Lekor

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Abstract: The common practice of mixing in keropok lekor processing is carried out continuously to produce a homogenize batter with no specific time and sequence established yet. The goal of the work was to study the effect of different mixing time and sequence (intermittent and continuous) against the physicochemical and sensory properties of keropok lekor. The keropok lekor batter was mixed for 6, 12 or 18 min non-stop for the continuous mixing, while the intermittent mixing was carried out for 6 min (2 min mix and 2 min break), 12 min (4 min mix and 2 min break), and 18 min (6 min mix and 2 min break). The intermittent mixing for 6 min was the best method among all the treatments, where the results for the moisture content (56.60±2.20), protein content (10.23±1.21), water holding capacity (83.33±11.02), linear expansion (5.86±2.32), cooking yield (107.15±0.60), lightness, redness and yellowness (colour), hardness, cohesiveness, springiness and chewiness (texture profile analysis) and sensory evaluation scores were equivalent ($p>0.05$) or better ($p<0.05$) than the continuous mixing or longer time of treatments. This indicates that shorter time of mixing (6 min) with intermittent mixing sequence is sufficient to produce a good quality and homogenized dough of keropok lekor.

Keyword: Food technology; mixing time; mixing sequence; continuous mixing; intermittent mixing; keropok lekor

1. Introduction

Keropok lekor also known as fish sausage or fish cracker (fish keropok) is a traditional snack made from minced fish flesh and mixed with starch flour, salt, crushed ice and monosodium glutamate [1]. Several types of fish that can be used in the production of keropok lekor such as '*Selayang*' (Trachurus trachurus), '*Tamban*' (Sardinella albella), '*Cencaru*' (Megalaspis cordyla) and others [2]. The quality of the keropok lekor can be preserved by using fresh fish as the main ingredient. [3]. In order to produce the best taste of keropok lekor, Malaysian Fisheries Department has prescribed that keropok lekor should have 30% fish content more than starch flour [4]. According to Food Regulations 1985, the protein must be 12% or above for un-fried fish keropok made from fresh fish, while, keropok produced from crustacean and mollusc must have 6.9% protein or above [5]. The quality of fish keropok depends on some factors such as the fish content and linear expansion [6]. The processing parameters such as the amount of leavening agents, variety types of flour, the proportion of flour to fish, homogenous dough (mixing), the thickness of the slices, the cooking oil temperature and the cooking time could influence the linear expansion of fish keropok [7]. A study reported that the fish keropok quality correlates with the percentage of linear expansion would affect the crispiness of the crackers [8].

A good mixing process could produce a very high quality of keropok lekor. Mixing is an important step during the production of a uniform keropok lekor with good expansion by homogenizing all the ingredients such as fish, starch, crushed ice and seasoning to the desired level[7, 9]. The problem that commonly occurs regarding keropok lekor production is the texture of keropok lekor which is tough and hardly to chew [9].

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Several studies showed that intermittent mixing was better compared to continuous mixing. For example, intermittent mixing resulted in better cured ham yield and quality compared to continuous mixing [10]. Intermittent mixing also with increased time improved migration of the cures (sodium chloride, dextrose, sodium nitrite) used in porcine muscle compared to continuous tumbling [11].

However, no information is currently available on the mixing time and mixing sequence (continuous and intermittent) on keropok lekor. This work was carried out to find out the effect of mixing time and sequence (intermittent and continuous) against the physicochemical and sensory properties of keropok lekor.

2. Materials and Methods

Sample preparation

Fresh fish “Kembung” (Rastrelliger kanagurta) and ‘*Selayang*’ (Trachurus trachurus) were obtained from a local supermarket. The unwanted parts like heads, bones and internal organs were removed from the fish and only the fish flesh was used. A fish meat separator machine (DH811-200, Taiwan) was used for the deboning process to produce minced fish.

Table 1 Formulation for keropok lekor.

Ingredients	Weight (%)
Minced fish flesh	50.3
Salt	1.26
Sugar	0.5
Tapioca starch	17.6
Sago starch	17.6
Monosodium glutamate	0.15
Crushed ice	12.6
Total	100

The formulation used for making the keropok lekor consisted of minced fish flesh, salt, sugar, tapioca starch, sago starch, monosodium glutamate (MSG) and crushed ice as shown in Table 1.

Mixing parameters

Mixing processes were carried out in a laboratory-scale bowl cutter (K3, Bench, Taiwan). The parameters were the mixing sequence and the mixing time. The mixing sequences used were continuous and intermittent. For the continuous method, all the ingredients were mixed together for 6, 12, and 18 minutes continuously. Whereas for the intermittent method, it was carried out for 6 min (2 min mix and 2 min break), 12 min (4 min mix and 2 min break), and 18 min (6 min mix and 2 min break). During the resting time of intermittent mixing, the ingredients added were separated into several stages. The first stage was mixing the fish flesh with salt, sugar, and MSG together, for the second stage, the sago and tapioca starch were added and the last stage the crushed ice was added. The experiments were carried out in triplicates.

Moisture and protein analysis

The samples was dried at 105℃ overnight to determine the moisture content, while Kjeldahl method was used to measure the crude protein according to AOAC methods [12].

Cooking yield

The amount of cooking yield was determined after the boiling process. The calculation used to calculate the amount of cooking yield is shown is Eq. 1

$$\text{Yield (\%)} = WAB/WBB \times 100 \qquad (1)$$

Where WAB is the weight after boiled and WBB is the weight before boiling [13].

Linear expansion

The linear expansion (%) was determined from the boiling process of keropok lekor. The length of the sample was calculated based on Eq. 2.

$$\text{LE (\%)} = (LAB - LBB)/ LBB \times 100 \qquad (2)$$

Where LE is the linear expansion, LAB is the length after boiled and LBB is the length before boiling [14].

Water holding capacity (WHC)

The water holding capacity determination was carried out based on Lin and Huang's method [15] but with minor adjustment. The homogenized raw keropok lekor (5 g) and distilled water (10 ml) were mixed in a centrifuge tube (50 ml) and centrifuged (Kubota 5800, Japan) for 10 min at 15℃ at 2,000 g. WHC was calculated based on the Eq. 3.

$$WHC = WBC - WAC / OSW \times 100 \quad (3)$$

Where WHC is the water holding capacity, WBC is the water before centrifuge, WAC is the water after centrifuge and OSW is the original sample weight.

Texture profile analysis (TPA)

Texture analyzer (TA-XT2i, Stable Micro System, UK), fitted with Warner-Bratzler blade and a 25kg load cell was used to determine the hardness, cohesiveness, springiness, and chewiness of the keropok lekor. The texture analyzer was set with pre-test speed at 2.0 mm/s; post-test speed at 2.0 mm/s; distance at 7.5 mm; and time at 5.0 s.

Color analysis

A calorimeter (Minolta spectrophotometer CM 3500d, Japan) was used to measure the colour of the cooked keropok lekor and was indicated by lightness level (L*), redness level (a*) and yellowness level (b*).

Sensory evaluation

The hedonic scaling by Maqsood et al. [16] with several modifications was used to evaluate the sensory attributes (appearance, colour, texture, chewiness, taste, and overall acceptability) of the keropok lekor. This evaluation involved 30 panellists. Each sample consisted of 2.0 cm of keropok lekor which was labelled with 3-digit random code.

Statistical analysis

Two-way analysis of variance (ANOVA) was used to analyze the data (Minitab 16 Inc. USA). The differences were considered significant at $p < 0.05$.

3. Results and Discussion

Moisture content, protein content, cooking yield, linear expansion and water holding capacity

The results of the moisture content, protein content, cooking yield, linear expansion and water holding capacity of the keropok lekor are shown in Table 2. Overall, there was a significant difference ($p<0.05$) between the time of mixing and mixing method for moisture content. The sample for 6 minutes of continuous mixing time was recorded to have the highest moisture content (59.50 %), which is significantly different ($p<0.05$) from the sample of keropok lekor mixing time more than 6 minutes. Keropok lekor that used the intermittent method of mixing was found to have significantly lower moisture content compared to the sample that used continuous mixing method. However, there were no significant differences recorded between the sample of keropok lekor that used continuous mixing with increasing of time for mixing.

The moisture content in keropok lekor might be related to the mixing time because the longer the mixing time might cause moisture loss from the keropok lekor. Haegens [17] stated that the amount of water inside a dough could be influenced by the mixing time, where longer mixing time hydrated the flour and other ingredients. Based on the result obtained, the highest moisture content was for samples that were mixed for 6 minutes for both continuous and intermittent mixing methods.

There was no significant difference ($p>0.05$) of protein content in keropok lekor between the time of mixing and mixing method for the intermittent mixing method. The results showed an increasing protein content for intermittent mixing method as the time of mixing increases. The highest amount of protein content was recorded by the sample of intermittent mixing with 18 minutes of mixing time (10.60%). There was a significant difference ($p<0.05$) between the percentages of protein content for sample of continuous mixing method as the mixing time increases.

Mixing process is carried out to produce a homogenous mixture and for protein formation. The protein content in keropok lekor was contributed by the fish flesh, sago starch, and tapioca starch [18]. The results showed that by using intermittent mixing method at 6 minute, the protein content was higher than the continuous method at similar time because during intermittent, the mixture was allowed to rest and the protein becomes easily to be extracted [19]. The high protein content could influence the binding property of the keropok lekor thus, improve its texture properties.

Table 2 Moisture, protein linear expansion, water holding capacity and cooking yield of keropok lekor.

Analysis	Time (min)	Type of mixing	
		Continuous	Inter-mittent
Moisture content (%)	6	59.50 ±1.36^{aA}	56.60 ±2.20^{aA}
	12	54.87 ±0.81^{bA}	56.20 ±0.72^{aA}
	18	55.07 ±0.50^{bA}	50.87 ±2.00^{bB}
Protein content (%)	6	9.57 ±1.51^{aA}	10.23 ±1.21^{aA}
	12	8.92 ±0.05^{aB}	10.40 ±0.81^{aA}
	18	10.80 ±1.51^{aA}	10.60 ±0.17^{aA}
Cooking yield (%)	6	109.01 ±1.44^{aA}	107.15 ±0.60^{aA}
	12	109.16 ±0.16^{aA}	105.21 ±0.53^{bB}
	18	105.42 ±1.15^{bA}	105.66 ±0.12^{bA}
Linear expansion (%)	6	7.35 ±1.80^{abA}	5.86 ±2.32^{aA}
	12	8.45 ±2.14^{aA}	3.00 ±0.60^{aB}
	18	3.17 ±1.85^{bA}	3.86 ±2.70^{aA}
Water holding capacity (%)	6	60.00 ±0.00^{Bb}	83.33 ±11.02^{aA}
	12	73.33 ±5.77^{abA}	61.33 ±2.31^{aB}
	18	79.67 ±8.96^{aA}	56.67 ±15.28^{aA}

Means ± SD with different superscripts capital letter (row-wise) () and small letter (column-wise) within the same analysis is significantly different ($p<0.05$).

A significant difference ($p<0.05$) was detected between the time of mixing and mixing method of the keropok lekor cooking yield. The data in Table 2 showed that the highest percentage of cooking yield was for keropok lekor mixed for 12 minutes with continuous mixing method (109.16%). Both results of continuous and intermittent mixing method showed significant differences ($p<0.05$) but the continuous mixing method was better compared to intermittent mixing.

The water holding capacity could be affected by the percentage of cooking yield of the keropok lekor. The cooking yield is as associated with the capability of the keropok lekor to hold the water. When there is any external pressure are applied onto it such as mixing, it would affect the cooking yield [20]. Longer mixing time could allow reduction of water content and might cause the cooking yield to be lowered.

The sample that has the highest expansion value was keropok lekor produced by using continuous mixing method for 12 minutes. The overall result showed that there was a significant difference ($p<0.05$) between the type of mixing method for linear expansion of keropok lekor. A research proved that poorly-expanded of keropok lekor was probably due to the insufficient mixing time [21]. The type of mixing affected the dispersion uniformity of the fish protein thus influenced the starch expansion. The fluctuation results obtained during the experiment might be contributed by the sequences of adding the ingredients during the production. The expansion of the keropok lekor also could be caused by the mixing, which is not homogenous thus decreased the gelatinization and lower the expansion ratio [22].

Significant differences ($p<0.05$) were determined between the samples and the highest value of WHC is for intermittent mixing method (6 minutes) which was 83.33%, while the increasing in mixing time caused the WHC in intermittent mixing method to be lower. For continuous mixing method, the highest WHC for 18 minutes was 79.67%. This indicates that the longer the mixing time with continuous mixing method, the higher the water holding capacity of keropok lekor.

Water holding capacity is significant in the gels and emulsions formation, which indicates the effectiveness of the keropok lekor in order to prevent the water loss. Mixing process promotes the aggregation of the protein network where it loses its structure thus results in the loss of WHC [23].

Texture Profile Analysis (TPA) and color

Table 3 showed the changes of hardness level when the keropok lekor are mixed with different time and mixing method. A significant difference ($p<0.05$) was determined for the interaction between mixing time and mixing method for continuous mixing method but no significant difference ($p>0.05$) for intermittent mixing method on the hardness of keropok lekor. From the data shown, the keropok lekor, which was mixed continuously for 18 minutes have the hardest texture compared to others.

The results were contrast compared to meat where the swelling of myofibrillar proteins in meat due to mixing may cause the reduction in hardness [24]. Nevertheless, from the result, the continuous mixing can be used to obtain tender texture of keropok lekor but the mixing time must be less than 18 minutes it can be considered as overmixing and gave the hardest texture.

A significant difference ($p<0.05$) was determined between the intermittent mixing method with the cohesiveness and the highest value of cohesiveness is produced at 18 minutes. For continuous mixing method, there was no significant different ($p>0.05$), which indicates that the continuous mixing did not give any affect towards the cohesiveness of the keropok lekor. As indicated in overall result, the mixing method did not have any significant difference ($p>0.05$) towards the cohesiveness but mixing time is shown to influence the cohesiveness of keropok lekor. The cohesiveness of keropok lekor or Malaysian fish sausages was in range of 0.28-0.42 [25]. Farouk et al. [26] stated that the cohesiveness of the cooked sausage was influenced by the sarcoplasmic and myofibrillar proteins. So, it can be concluded that the keropok lekor that had been produced by continuous mixing method is more acceptable for the cohesiveness value compared to the intermittent mixing.

There is no significant difference ($p>0.05$) between the springiness of keropok lekor for the mixing time and different mixing method.

Table 3 Texture and colour analysis of keropok lekor.

Analysis	Time (min)	Type of mixing	
		Continuous	Inter-mittent
Hardness (g)	6	2946.3 ±123.3^{bB}	3296.1 ±73.4^{aA}
	12	2518.2 ±292.7^{bB}	3335.4 ±253.2^{aA}
	18	3761.2 ±459.7^{aA}	3078.6 ±511.1^{aA}
Cohesive-ness (%)	6	0.369 ±0.02^{aA}	0.477 ±0.12^{abA}
	12	0.392 ±0.07^{aA}	0.282 ±0.15^{bA}
	18	0.442 ±0.13^{aA}	0.667 ±0.04^{aA}
Springi-ness (%)	6	0.79 ±0.05^{aB}	0.94 ±0.07^{aA}
	12	0.57 ±0.12^{aA}	1.34 ±0.85^{aA}
	18	0.81 ±0.17^{aA}	0.85 ±0.05^{aA}
Chewi-ness (J)	6	857.5 ±121.3^{aA}	1487.7 ±432.3^{aA}
	12	588.2 ±252.1^{aA}	970.0 ±222.8^{aA}
	18	1454.6 ±919.9^{aA}	1747.6 ±265.4^{aA}
Lightness (L*)	6	62.61 ±3.16^{aA}	59.72 ±6.21^{aA}
	12	55.81 ±2.78^{aA}	57.00 ±5.10^{aA}
	18	55.79 ±4.11^{aA}	56.19 ±2.86^{aA}
Redness (a*)	6	2.38 ±0.15^{aA}	2.69 ±0.22^{aA}
	12	1.95 ±0.23^{aA}	2.01 ±0.43^{abA}
	18	2.10 ±0.23^{aA}	1.47 ±0.31^{bB}
Yellow-ness (b*)	6	11.61 ±1.60^{aA}	10.49 ±2.45^{aA}
	12	8.64 ±1.57^{abA}	8.18 ±2.21^{aA}
	18	7.98 ±0.24^{bA}	7.31 ±0.80^{aA}

Means ± SD with different superscripts capital letter (row-wise) () and small letter (column-wise) within the same analysis is significantly different ($p<0.05$).

Springiness can be considered as the tendency of the keropok lekor to return into original shape when a force applied to it. The springiness also can be influenced by the quality of the protein [23]. The springiness values obtained were not too high might be due to the protein network of the dough during mixing process was not well developed or the speed of the mixing was insufficient to produce the good springiness of keropok lekor.

The overall result of chewiness showed that there was no significant difference ($p>0.05$) for both continuous and intermittent mixing methods towards the chewiness of keropok lekor. The highest chewiness was recorded for intermittent mixing method at 18 minutes. Pietrasik [27] reported that protein content was the major factor that influences the chewiness. So that, the mixing time can be one of the factors that can encourage the chewiness since the mixing process promotes the interaction of the protein network in keropok lekor.

The color of keropok lekor is presented in Table 3. The overall results showed no significant difference ($p>0.05$) between the mixing time and mixing method towards the lightness (L*). The highest lightness value is shown by the keropok lekor with continuous mixing method for 6 minutes. However, for intermittent mixing method, the value of the lightness decreases when longer mixing time used. Therefore, for both continuous and intermittent mixing method, the values of the L* are still between the range, which is acceptable for the consumers. A study by Huda et al. [25] on the Malaysian commercial fish sausages resulted with the L*, a* and b* were from 58.73-79.56, -0.58-17.43 and 12.69-22.96, respectively.

The value of redness (a*) of keropok lekor decreases significantly ($p<0.05$) for the intermittent mixing method whereas for continuous mixing method, there is no significant difference between the sample. The overall showed that there is a significant difference ($p<0.05$) between the mixing time with the redness of keropok lekor while the mixing method did not affect the redness of the keropok lekor. All the values of a* are acceptable as the color of keropok lekor Huda [25].

The average range of yellowness (b*) for keropok lekor for continuous mixing method was from 7.98 to 11.61 and for intermittent mixing method was 7.31 to 10.49 which are not in the range that fulfils the characteristics of the Malaysian fish sausages. The whole result showed that the change in mixing time have the significant effect on the yellowness of the keropok lekor. The yellowness decreases for both continuous and intermittent mixing method. However, there is no significant difference ($p>0.05$) between the intermittent mixing method with the yellowness of keropok lekor. The result showed that the longer the mixing time, the lower the yellowness of keropok lekor.

Sensory evaluation

There is no significant difference ($p>0.05$) for the appearance, color, taste and overall acceptability of the keropok lekor (Table 4). This indicates that mixing method and mixing time did not affect the acceptability of consumers based on those attributes. However, for the texture and chewiness, there is a significant difference ($p<0.05$) which indicates the mixing time and mixing method gave the different texture and chewiness to the keropok lekor.

4. Conclusion

The result from the study shows that keropok lekor that have been produced using intermittent mixing method for 6 minutes showed the best result according to the physicochemical properties and sensory evaluation. From the experiment, keropok lekor that was produced by intermittent mixing method was having higher protein content, better water holding capacity, a better result for texture and colour, and the most preferable for the sensory evaluation. This indicates that 6 minutes is enough to homogenize the dough by having the resting time and loading the ingredients by stages. Thus, better texture and quality of keropok lekor can be produced. This new finding might be useful to help the producer of keropok lekor in order to optimize their production time and cost.

Table 4 Sensory evaluation of keropok lekor for continuous and intermittent mixing method

Sample	Appearance	Color	Texture	Chewiness	Taste	Overall acceptability
Continuous 6	5.83 ± 2.17[a]	5.67 ± 1.73[a]	5.70 ± 1.58[ab]	5.83 ± 1.62[ab]	6.13 ± 1.55[a]	5.97 ± 1.61[a]
Continuous 12	6.23 ± 1.48[a]	6.20 ± 1.61[a]	6.13 ± 1.28[a]	5.87 ± 1.57[ab]	6.10 ± 1.86[a]	6.00 ± 1.51[a]
Continuous 18	5.90 ± 1.37[a]	5.97 ± 1.40[a]	4.93 ± 1.51[b]	5.03 ± 1.45[b]	5.73 ± 1.55[a]	5.47 ± 1.46[a]
Intermittent 6	6.27 ± 1.48[a]	6.27 ± 1.64[a]	6.17 ± 1.56[a]	6.20 ± 1.47[a]	6.13 ± 1.63[a]	6.33 ± 1.65[a]
Intermittent 12	6.43 ± 1.50[a]	6.27 ± 1.57[a]	5.77 ± 1.46[ab]	6.00 ± 1.44[ab]	5.80 ± 1.63[a]	6.00 ± 1.53[a]
Intermittent 18	6.43 ± 1.65[a]	5.80 ± 1.69[a]	5.30 ± 1.47[ab]	5.27 ± 1.51[ab]	5.83 ± 1.29[a]	5.60 ± 1.25[a]

Means that do not share same letter are significantly different ($p<0.05$) column-wise.

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